

A Comparative Evaluation of Antimicrobial Effect of Snuhi-Apamarg-Ksharsutra and Udumbara-Ksheer Sutra in the Management of Bhagandara (Fistula-In-Ano)

Research Article

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Abstract

Bhagandara one of the eight grave disorders, pertaining the perianal region is arduous to treat, has been described in detail by Acharya Sushruta. The Ayurvedic description of Bhagandara can be identified as associated with an anal fistula. An anal fistula is defined as an internal opening in the altered skin or mucosa of the anal canal or rectum and an exterior opening in the perianal skin region. Among the various modalities available for the treatment of bhagandara, ksharasutra is described as gold standards for treatment and cure. The ksharasutra possess antimicrobial properties and can be established as first line of treatment for bhagandar(fistula-in-ano). The modern approach for fistula-in-ano is surgical procedure. Antibiotics are commonly used along with surgery for the treatment of fistula-in-ano. The aim of this study was to comparatively evaluate the antimicrobial properties amongst Snuhi-apamarga kshar sutra and Udumbara ksheer sutra. A total no. of 60 participants were enrolled as per the criteria for the study and pus samples were collected as per the standard operating procedure. Samples of Snuhi-apamarga kshar sutra and Udumbar ksheer sutra were already given in laboratory for testing the sensitivity. Reports were collected and analyzed on different parameters and conclusion was drawn accordingly. It was concluded that gram positive monomicrobial infections were more common especially involving staphylococci. Also, in the culture and sensitivity test Snuhi- apamarga ksharsutra was found to be more effective in comparison to Udumbara ksheer sutra as the microbes were highly sensitive to the Snuhi Apamarga kshar sutra. This can be contributed to the multiple contents in Snuhi Apamarga kshar sutra.

Keywords: *Ksharsutra* , *Bhagandara*, *Udumbara ksheer*, Microbes, *Fistula-in-ano*.

Introduction

Ayurveda incorporating the eight folds of Kaya, bala, graha, urdhavanga, shalya, danshtra, jara, vrisha has held itself time tested and the most scientific medical science since ancient times. Surgery (shalyatantra) was at its pinnacle during the time of Acharya Sushruta, who has been aptly called as the 'Father of Modern Surgery'. He described 'Astomahagada' (Eight Grave Disorders). Bhagandara (Fistula-in-ano) being one of them, it is said extremely difficult to be cured. (1) The disease in which there is 'darana' i.e Splitting of 'bhaga' perianal region is 'bhagandara'. It begins with a Pidika (boil) in perianal region, which gets suppurated and bursts to form Bhagandara. (2) A variety of therapeutic approaches for

the treatment of Bhagandara has been prescribed by Acharya Sushruta, including oral drugs, local applications, surgical operations, and para-surgical intervention. Currently, Ksharsutra therapy is the most approachable and appealing treatment technique among para-surgical procedures for anorectal fistula.(3) According to a recent study on the prevalence of anal fistula in India by the Indian Proctology Society, this disorder among the anorectal disorders affected roughly 17 to 20% of the population in different states, and 4% of new patients were reported to have it. (4) According to modern medical science similar features of bhagandar can be correlated with Fistula-in-ano. An improper passage between two epithelial surfaces is referred to as a fistula in ano, and the track is typically coated by unhealthy granulation tissues. The main cause known for fistula in ano is crypto glandular infection of anal crypts. (5).

Another point about Bhagandara (fistula-in-ano) which needs to be given attention is "Infection". Most of the time fistula-in-ano is a sequel of perianal abscess which is an infectious condition. Also due to the particular site of this fistula and direct connection with

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the anal canal, the track is prone to get infected again and again. In Ayurvedic text also (in the context of Unmargi Bhagandara) Sushruta mentioned that Krimi's (microbes) appears in the wound in at Guda (anal) region, which destroy away the rectum (anus) and produces tear at many sites. (6) Charaka also enumerated 'Krimi' to be a primitive cause of Bhagandara (7) So, the fistula tract is always loaded with various microorganisms, which increases pus discharge and interferes with the healing mechanism. Of all the modern surgical and Para surgical modalities available for the treatment of fistula-in-ano (bhagandara) antibiotics form a mandatory prescription for the post-operative management. It is a serious concern today that bacteria are growing increasingly resistant to these medications and that they have certain unfavorable side effects. Now, the drugs which are used in Ksharsutra preparation are having "Krimighna" (antimicrobial) properties. (8) So, it is reasonable that Ksharsutra may possess some anti-bacterial properties, which help in controlling infection of fistula tract, thus promoting healing. (9) This property has not been studied or proven with modern parameters till today. Thus, in present study Krimighna (antimicrobial) property of Snuhi-Apamarg-Ksharsutra and Udumbara Ksheersutra is assessed in terms of modern investigation method (i.e. Culture and Sensitivity test).

Aims and Objectives

To do a comparative study of antimicrobial property of Snuhi-apamarg-ksharsutra and Udumbara-ksheer sutra. Also, to find out most commonly present bacteria in the fistula tract.

Methodology

Patients having the classical symptoms of bhagandar were selected for the study.

- Study Design - Randomized control trial. It was a Single blind trial.
- Source of Data – The patients enrolled in the Shalyatantra OPD were enrolled for the study based on examination method as per the protocol.
- Research Proforma: A detailed case paper proforma was prepared for the present study incorporating all the relevant point from both the Ayurvedic as well as modern point of view.

Diagnostic criteria

Patients were selected on the basis of thorough local and systemic examination, history taking and investigations; as per the case paper proforma specially prepared for the study.

Criteria for enrolment – Local examination, Per rectal examination, Probing, and Proctoscopy.

Inclusion criteria

- Age group between 20yr-60yr age.
- Low anal uncomplicated fistula.
- Fistula having less than three external openings.
- Irrespective of sex, economic status, religion, occupation

Exclusion criteria

- Fistula connected with other organs like urethra, vagina etc.
- Patients with systemic diseases like T.B., D.M., and Cardiac disease.
- Patients with CA Rectum, Crohn's disease, Ulcerative Colitis.
- Patients with immune compromised diseases like HIV, HBsAg.
- Patients with other malignancies.

Ksharasutra

Contents and Equipment required for the Ksharasutra preparation were:

1. Snuhi Apamarga Ksharsutra
 - a.) Barbour's linen thread No.20
 - b.) Snuhi Ksheer (Euphorbia nerrifolia Linn.)
 - c.) Apamarga Kshar (Acchyranthus aspera Linn.)
 - d.) Haridra Powder (Curcuma longa Linn.)
2. Udumbara Ksheersutra (Ficus racemose Linn.)
 - a.) Barbour's linen thread No.20.
 - b.) Udumbara Ksheer
3. Ksharsutra Hangers
4. Ksharsutra Cabinet WITH UV light
5. Sterilized Test tube.
6. Gauze piece and Gloves.

Method of Ksharasutra preparation

Required raw drugs collection and preparation of Snuhi-apamarga ksharsutra and Udumbara ksheer sutra was done as per standard Protocol. (10),(11)

Investigations

Routine investigations were done:

- Hematology: - Hb%, CT, BT, TLC, DLC, ESR, BSL, HIV, HBsAg, Sr. Creatinine
- Urine- Routine and Microscopic
- Specific investigations (if required)
- Fistulogram
- X ray chest (PA view)
- X ray pelvis with both hip joints
- Barium enema
- Biopsy of fistulous track in suspected cases of tubercular fistula
- Mountox test and IgG
- Sigmoidoscopy, Colonoscopy

Culture and sensitivity test

Before starting the treatment, the pus smear was taken from fistulous track opening and was sent to pathologist for studying its culture and sensitivity with antibiotics as well as with the contents of Ksharasutra.

Method of culture and sensitivity test

The culture and sensitivity tests were carried out as per the standard method as follows:

Sample collection

Procedure of pus collection from fistula for the isolation of microorganisms

- By sterile technique, pus from the abscess was collected at the time of incision and drainage

procedure. 1 to 4 ml of pus was collected or aspirated, in a sterile syringe.

- If the pus was not being discharged, the sterile cotton wool swab was used to take sample from the depth of external opening of fistula track.
- Care was taken to avoid contamination of sample with normal flora.
- The collected sample was labelled and sent to the laboratory immediately.

Figure 1: Pus Sample Taken For Culture And Sensitivity Test



Testing (fig. no. 2, 3)

Testing was done in the pathology laboratory as per the standard protocol-

1. Pus culture and identification of pathogenic organism

Pus samples were inoculated on Mueller Hinton at 37°C temperatures for 12 to 24 hours. Then colonies were identified using standard biochemical reactions.

2. Antibiotic sensitivity

In the positive culture, this test was carried out using Kirby-Bauer disc diffusion method on Mueller Hinton agar plate. In this method filter paper discs were used, 6.0mm in diameter, charged with the concentrations of the drugs. The discs were dried and stored in the refrigerator. A suitable dilution of a broth culture was spread on Muller Hinton agar plate using sterile swabs. After drying the plate at 37°C for 30 minutes, antibiotic discs were applied with sterile forceps. After overnight incubation, the degree of sensitivity was determined by measuring the zones of inhibition of growth around the discs. Growth was inhibited around discs containing antimicrobials to which the bacterium was susceptible but not around those to which it was resistant.

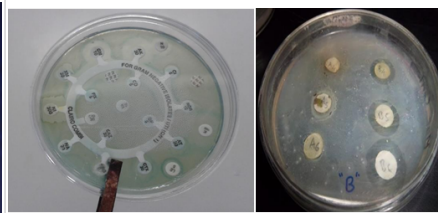
3. Ksharsutra sensitivity

This was done by the same method as for testing antibiotic sensitivity; only in place of antibiotic discs, the Ksharsutra/Ksheersutra contents discs were used. In this method filter paper discs were used, 6.0mm in diameter, charged with the concentrations of the Ksharsutra/Ksheersutra contents. The discs were dried and stored in the refrigerator. A suitable dilution of a broth culture was spread on Muller Hinton agar plate using sterile swabs. After drying the plate at 37°C for 30 minutes, ksharsutra discs were applied with sterile forceps. After overnight incubation, the degree of sensitivity was determined by measuring the zones of inhibition of growth around the discs. The zones of inhibition of growth around disc were measured and degree of sensitivity determined in same manner as above.

Figure 2: K.B Disc, to check sensitivity



Figure 3: Antibiotic and Ksharasutra sensitivity (Zone of inhibition of microbial growth can be seen around the discs)



Reports

The reports of culture and sensitivity tests were obtained after 2 to 3 days, after sending of sample. Negative cultures were also reported in 1 to 2 days.

Assessment criteria

Culture and Sensitivity test reports

After studying the zones of inhibition of growth around disc, the degree of sensitivity was measured and interpretation was mentioned as follows, by pathologist. (For both types of reports i.e. Antibiotics as well as Ksharsutra contents.)

Table 1: Showing grading of ‘Sensitivity’

Score	Sensitivity
+	Mild/sluggish sensitive
++	Moderately sensitive
+++	Highly sensitive

Statistical Analysis

For culture sensitivity test analysis: - percentage data was considered.

Observations and Results of Culture and Sensitivity test

Table 2: Showing results according to ‘culture’

Sr. no.	Culture for organism	No. of pts	%
1	Positive	53	88.33%
2	Negative	7	11.67%
	Total		100%

It was observed that out of 60 patients included in study, 88.33% patients had cultures positive for microorganism; while 11.66% patients pus culture were negative for any septicemic condition.

Table 3: Showing results according to ‘type of organism’

Sr. no.	Type of organism	No. of pts	%
1	Gram positive Bacteria	32	53.33%
2	Gram negative Bacteria	21	35%
3	Negative culture	7	11.67%

Out of the total cultures; Gram positive bacteria was identified in 53.33% patient’s pus culture, while gram negative bacteria were identified in 35% cultures.

Table 4: Showing result according to identified causative organism

Sr. no	Organism	No. of pts	%
1	GM +VE STAPHYLOCOCCI	29	48.33%
2	GM +VE STREPTOCOCCI	3	15%
3	GM –VE STAPHYLOCOCCI	9	5%
4	GM –VE PSEUDOMONAS	8	13.3%
5	GM –VE KLEBSIELLA	4	6.67%
6	NEGATIVE CULTURE	7	11.67%
	Total	60	100%

It was observed that, the positive cultures revealed monomicrobial isolates. In maximum cultures i.e. in 48.33% GM +ve staphylococci was isolated predominantly; Followed by GM +ve streptococci and GM –ve pseudomonas in 15% and 13.3% cultures respectively. GM –ve staphylococci was identified in 5% and GM –ve klebsiella in 6.66% cultures.

Table 5: Showing results according to ‘Sensitivity with Ksharsutra’

Sr. no.	Sensitivity to	Sensitivity grading			Total	% (out of 60 pts)
		High (+++)	Moderate (++)	Mild (+)		
1	Snuhi+Apamarg + Haridra	21	1	4	26	43.33%
2	Snuhi Ksheer	9	3	7	19	31.66%
3	Apamarg Kshar	16	2	2	20	33.33%
4	Haridra Churna	7	12	7	26	43.33%
5	Udumbara Ksheer	1	4	4	9	15%

From above table it can be observed that cultured organisms were mostly sensitive to combination of Snuhi+Apamarg+Haridra. Out of 60 reports 26 reports i.e. 43.33% showed sensitivity of Snuhi-Apamarg-Haridra Ksharsutra; out of them 21 were highly sensitive while 1 and 4 were moderately and mildly sensitive respectively. In 26 i.e. 43.33% reports sensitivity was shown to Haridra Churna (7 highly, 12 moderately and 7 mildly sensitive). Then 33.33% showed sensitivity to Apamarg Kshar (out of 20; 16 highly, 2 moderately and 2 mild sensitive). In 31.66% cases pathogen was sensitive to Snuhi Ksheer (out of 19; 9 highly, 3 moderately and 7 mild sensitive). In very few cases i.e. 15% sensitivity was observed to Udumbara Ksheer (1 highly, 4 moderately, 4 mildly sensitive out of 9).

Discussion

Ano-rectal disorders like hemorrhoids, fissure, fistula etc. are progressively increasing in society. Some important causes are sedentary life style, irregular and inappropriate diet, prolonged sitting and psychological disturbances like anxiety and depression etc. These also

disturb the metabolic activities and digestion, thus leading to constipation.

Constipation is the root cause of all ano-rectal disorders. Fistula in ano is one of the most common and notorious disease among all ano-rectal disorders which produces inconvenience in routine life. Literary simulation of bhagandara coincided with the description of fistula in ano as described in modern literature. Its description is available in Brihatrayi as well as in Laghutrayi. Ayurvedic Kshara Sutra has been proved as a big revolution in the treatment of fistula in ano. Through continuous researches and experiments, a standard Ksharsutra was established which is mostly in use for the bhagandara treatment.

Antimicrobial effect

Conventionally prepared Ksharasutra is well known for its therapeutic effect in the anorectal diseases like Bhagandara. Many clinical studies are being carried out with variations in the Kshara and the latex in order to find out a better alternative to conventional Snuhi Apamarg Ksharsutra. In present study Udumbara Ksheer was considered as alternative to Snuhi Ksheer.

These herbs applied to the thread are having good antimicrobial activity and healing ability, which is expected in the treatment of the diseases. The study was aimed to evaluate the anti-microbial properties of ksharsutra in culture techniques. And, also to find out recent trend of bacteria present in fistula track.

The results obtained from the study are represented in percentage data-

- Out of 60 culture reports, 88.33% patients had cultures positive for pathogenic organism; while 11.66% pus culture were negative for any septicemic condition (Table no. 2).
- No doubt fistula in ano is an infectious condition; the anorectal canal harbours infection from the colon every now and then. Reason behind the few negative cultures may be the self-medication of some patients with antibiotics, before attending the hospital; which makes discharge from track sterile temporarily.
- Out of the total cultures; Gram positive bacteria was identified in 53.33% patient’s pus culture, while gram negative bacteria were identified in 35% cultures. (Table no. 3)

This shows, as per present study, there is more incidence of Gram-positive bacteria in fistula in ano. Compared with gram-positive bacteria, Gram-negative bacteria are more resistant against antibiotics because of their impenetrable cell wall. 90-95% of gram-negative bacteria are pathogenic; on other hand, many Gram-positive bacteria are non-pathogenic.

This can explain that antibiotics are not really required during treatment with Ksharsutra as mostly track is infected with Gram positive bacteria which are mostly non-pathogenic and also as drugs used in Ksharsutra are having antimicrobial activity.

From the positive cultures, it is seen that all revealed monomicrobial isolates. Polymicrobial infection was not seen in any of the case under study. In maximum cultures (48.33%) incidence of GM +ve

staphylococci was found; Followed by GM+ve streptococci and GM -ve pseudomonas in 15% and 13.3% cultures respectively. GM -ve staphylococci was identified in 5% and GM -ve klebsiella in 6.66% cultures. (Table no. 4)

From the study four different species of bacteria were isolated from the 60 pus samples obtained from fistula in ano patient. All of them in the form of monomicrobial infection. Reason behind not getting polymicrobial isolates may be that patients who are immune- compromised or with diabetes, tuberculosis etc. were excluded from the study, which are more prone to get infected because of increased susceptibility to bacterial infection thereby encouraging their colonization.

Predominantly found staphylococci are Gram positive cocci which are ubiquitous and are the most common cause of localized suppurative lesions in humans. They are the primary parasites colonizing skin, skin gland and mucous membranes. Coagulase negative staphylococci and streptococci also constitute a major component of normal flora of body. Klebsiella is the second most (after Escherichia coli) populous member of the aerobic bacterial flora of intestine. Pseudomonas is also a ubiquitous found in water, soil and moist environments; and typically, opportunistic in causing infection. It is also a part of resident aerobic bacterial flora of colon. So, among isolates; staphylococci and streptococci are skin derived organism; whereas Klebsiella and pseudomonas are gastrointestinal track derived organisms.

Considering the crypto-glandular theory of aetiopathogenesis, it is obvious to expect to get bacteria of intestinal flora in pus culture of fistula -in ano. Though Klebsiella and pseudomonas found in few cases; E.coli was not observed in any of the case.

From analysis of antimicrobial activity of Ksharsutra, which was one of the main aims of the study, it is observed that cultured organisms were mostly sensitive to combination of Snuhi+Apamarg+Haridra (43.33%). Considering the intensity, most cases showed high sensitivity (21 reports out of 26). Considering the individual contents of Ksharsutra, Sensitivity to Haridra churna was predominant (43.33%); most of them moderate sensitive (in 12 reports out of 26). Another content Apamarg Kshar showed sensitivity in 33.33%; most of them of highly sensitive grade (in 16 reports out of 20). The main content Snuhi Ksheer showed minimum sensitivity out of three contents of Snuhi Apamarg Ksharsutra (31.66%); mostly highly sensitive (9 reports out of 19). In very few cases bacteria showed sensitivity to the content of experimental group which was Udumbara Ksheer (15%); that too mild and moderate sensitivity (4 reports each, out of 9). (Table no. 5)

This proves the “krimighna” property or antimicrobial action of Ayurvedic Ksharsutra. It can be said that Ksharsutra used currently are active against all commonly found organisms of fistula-in-ano track.

Out of two types of Ksharsutra considered for the study; Snuhi Apamarg Ksharsutra is almost three times better than Udumbara Ksheersutra in terms of

antimicrobial activity. But from above observation it can be said that antimicrobial activity of Snuhi Apamarg Ksharsutra is mostly contributed by Haridra (Turmeric) and partly by rest two contents.

As mentioned in “drug review” Haridra is described as Krumighna (12) or Krumihara (13). Apamarg is included in “Krumighna” Gana by Charaka(14); and Sushruta included it in “Arkadi” Gana which is mentioned to have “Krumisadan” property. Also, in description of Kshara, Sushruta mentioned that it works against Krumi (Su.Su. 11/5)(15). Snuhi Ksheer is not mentioned as “krimighna” in Ayurvedic text; but its Katu Rasa and Laghu-Tikshna Guna can contribute to this effect. Udumbara is also not described as “Krumighna”; but from its Kashay Rasa and Ruksha Guna can act against Krumi.(16)

“Haridra” *Curcuma longa* linn. is well proven for its antimicrobial as well as antifungal and antiviral action. In one research, as mentioned in drug review, it was observed that the ethanolic extract of the leaves and stem of the *Acchyranthus aspera* plant inhibited the growth of *Bacillus subtilis* and *Staphylococcus aureus* bacterial strains. (17)

“PH” plays an important role in this antimicrobial action. The majority of pathogenic bacteria grow best at neutral or slightly alkaline reactions (PH 7.2-7.6). The PH of Snuhi Apamarg Ksharsutra (9.3) which is more alkaline; as well as Udumbara ksheersutra (5.4) which is acidic is not favorable for growth of the organisms. (18)

Infection is one factor which retards healing and in case of fistula-in-ano, the infection cannot be eradicated by any amount of antibiotic and antiseptic measures. The Ksharsutra prepared from above mentioned drugs helps to fight with infectious condition of fistula track as it combat with micro-organism and also helps to get rid of secondary infection. Ultimately it has a role in healing of fistula track.(19)

Conclusion

From “Culture and sensitivity” study, the observed pattern of pathogens involved in fistula-in-ano is that – There is more incidence of monomicrobial infection with Gram positive bacteria.

As per the study, Predominant bacterial species involved in infection of fistula track is – “staphylococci”.

From culture and sensitivity with Ksharsutra, it is concluded that Ksharsutra is having antimicrobial effect which helps to promote healing of fistula track.

The antimicrobial effect is seen more in Snuhi apamarg Ksharsutra as there is combine effect of three drugs, with Haridra playing the major role. Comparatively antimicrobial property is less in Udumbara Ksheersutra.

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